Pesticides and Bees[1]

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ABSTRACT Alfalfa leafcutting bees, *Megachile rotundata* (F.), and alkali bees, *Nomia melanderi* Cockerell, are inherently more tolerant than honey bees, *Apis mellifera* L., to most insecticides. However, their greater surface-to-volume ratios cause them to be more susceptible to most field-weathered residues. Insecticides are minimally hazardous to all bees through nectar contamination, and to the alfalfa leafcutting bee via contamination of pollen-nectar stores and leaf pieces. Both acidifiers and acaricides can increase the hazard of insecticide combinations to all three species of bees. Most herbicides and all fungicides are essentially nontoxic to bees. Wild bees often exhibit different poisoning symptoms than honey bees. This is especially notable with exposure to carbaryl.

We have conducted many investigations of the effects of pesticides on several species of bees over the past 30 years. We have looked at the special hazards of certain insecticide formulations, the foraging behavior of bees, the effects of environmental conditions, and other factors involved in bee poisoning problems. Here, we review the major conclusions developed from these studies and provide brief descriptions of testing procedures evolved through these investigations.

The alfalfa leafcutting bee, *Megachile rotundata* (F.), has a greater inherent tolerance to most chemicals in the laboratory than either the honey bee, *Apis mellifera* L., or the alkali bee, *Nomia melanderi* Cockerell. In one of the few published articles on comparative toxicology of the leafcutting bee, the alkali bee, and the honey bee, Torchio (1973) found that the leafcutting bee was least susceptible to 73% of the materials tested and the alkali bee was less susceptible than the honey bee to 79%. Ahmad and Johansen (1973) looked at enzyme systems and other possible sources of the variation in tolerance. The only positive correlation found was with higher pH of the body fluid in the leafcutting bee.

In what appears to be an anomaly, the inherent insecticide tolerance of the leafcutting bee is reversed under field conditions. Since essentially all cases of poisoning occur from chance adherence of insecticidal residues to bees foraging treated plants, Johansen (1972a) suggested that the main reason for susceptibility in the leafcutting bee was its greater surface-to-volume ratio. This became more convincing when we made detailed measurements of the three species in combination with published diagrams and fresh weights. Analyses provided the following data for the leafcutting bee (LB) female, the alkali bee (AB) female and the honey bee (HB) worker. The surface-to-volume ratios (mm^2/λ) were: LB, 94/33; AB, 165/87, and HB, 186/128. The index for LB, AB, and HB was 2.0, 1.3, and 1.0, respectively.

Materials and Methods

Residue Exposure

Toxicity of field-weathered pesticide residues is assessed by treating 0.004-ha plots of second-growth alfalfa. Applications are made with a hand sprayer at a pressure of 1,760 g/cm^2, delivering 38.3 liters/ha. Alfalfa foliage samples (upper 15-cm portions of plants) are clipped into 2.5- to 5-cm lengths, and about 500 cm^3 is placed in each cage. Residual test exposures are replicated four times by caging 60 to 100 worker honey bees, 20 to 40 leafcutting bees, and 12 to 18 alkali bees with each of four foliage samples per treatment and time interval.

Honey bee workers are obtained from top supers of colonies and anesthetized with CO_2 to facilitate handling. Leafcutting bee and alkali bee prepupae in leaf piece cells and soil cores, respectively, are incubated at 29.5 to 31°C and 60% relative humidity (RH). Emerging adults are trapped in canisters fitted with screen funnels and chilled in a refrigerator at 1.5°C to facilitate handling. It is appropriate to compare predominantly 3-week-old honey bee workers with 1-day-old alfalfa leafcutting bees and alkali bees in these studies, because these are the ages of each species as it commences field activity. Whereas leafcutting bees becoming increasingly susceptible to insecticides with age, newly emerged honey workers are particularly susceptible and become more tolerant as they approach middle age. Various kinds of cages have been used since testing was initiated in 1953, but we standardized with a 15-cm plastic petri dish model. A strip of metal screen (6.7 meshes per cm) 45 cm long and 5 cm wide is stapled to form a circular insert which provides ample room in the cage for honey bees to fly and to defecate (essential for survival). Syrup prepared from either 50% sucrose or 50% honey and water is fed in a wad of cotton (5 by 5 cm), and the bees are held at 29.5 to 31°C and 60% RH for 24-, 48-, and 72-hour mortality counts.

Nectar Contamination and Leaf Piece Contamination

Biennial white sweet clover, *Melilotus alba*, was selected for nectar contamination studies because it is highly attractive to bees and easily adapted for greenhouse cul-

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ture. After the initial growing season, roots of the plants are harvested in mid-December to ensure vernalization. Soil is removed by washing, and the roots are air dried. Captan is dusted on them to inhibit mold problems and they are stored in a cold room (2°C) in plastic bags.

Flowering is initiated by planting each root in a 15-cm pot and placing them in a greenhouse with 16-h day length, 27°C day temperature, and 16°C night temperature. When the roots are held in cold storage for at least 2 months, blooms are obtained 8 weeks after planting the roots and good numbers of flowers are produced.

Tests are mainly conducted in cages 81 by 61 by 91 cm with both alkali bees and alfalfa leafcutting bees. However, cages as small as 30 by 30 by 45 cm are large enough for leafcutting bee studies.

Similar methods are used for leaf piece contamination studies, except that nest blocks are added so leafcutting bee females are encouraged to construct cells. Untreated pollen and nectar plants and honey syrup are provided, whereas treated plants without blooms are the only source of leaf pieces.

Acute Oral

Predetermined microliter amounts of the pesticide in 50% honey syrup are injected into 6- to 10-mm lengths of 2.9-mm (ID) tubing fitted snugly into plastic snap caps. Test bees inactivated by chilling are placed singly into 12-ml glass bottles, the snap caps are attached, and each of 10 units with a given test dosage is placed horizontally in a 15-cm petri dish. Each set of feeding units is placed in a holding room at 29.5°C and 60% RH for 3 h (bees normally consume the treated syrup within 1 h).

After the feeding period, each group of 10 bees is placed in the standard holding cage (15 cm in diameter by 5 cm high) and fed 50% honey syrup. Mortality counts are taken at 24, 48, and 72 h.

Field Studies

Field tests of bee poisoning are conducted in the Pullman, Wash., area on 0.4-ha plots of alfalfa. Growers are asked to leave the small plot near the center of each test field when they cut their hay. Such isolated plots, 3.2 to 8 km apart and surrounded by hay and grain stubble force good visitation by honey bees. Two colonies with Todd dead bee traps (Atkins et al. 1970) attached are placed next to each plot after the alfalfa is in full bloom.

Recently, we have obtained good isolation and bee foraging activity by treating center-pivot irrigation circles of corn near Prosser, Wash. The circles were 52.7 ha each, were surrounded by non-bee crops, and were at least 11.3 km from other irrigated fields. We placed 8 to 10 colonies of honey bees with Todd traps attached next to each circular plot.

Wild bees are much less amenable to field tests of poisoning, but we routinely make observations and take samples at adjacent alkali bee nest sites and leafcutting bee shelters when insecticide trials are applied to 8.1- to 12.2-ha alfalfa seed fields. We have also established small leafcutter shelters with 200-tunnel nest blocks either on alfalfa seed plots covered by cages 6.1 by 6.1 by 1.8 m, or on isolated 0.1-ha alfalfa hay plots.

Calibration of Test Results

Based on a combination of laboratory and field experience, we have established hazard levels for ranges of mortalities obtained in residue exposure tests. These can be related to the numbers of dead bees obtained in Todd traps in field studies, and trap catches, in turn, can be related to the honey bee field force and total number of bees in the colony (Table 1).

Relationships between test mortalities, dead bee trap catches, and total populations can be most accurately determined with honey bees because colony population data are available. However, proportions of populations affected in relation to test mortalities can probably be translated to wild bee species to obtain reasonably accurate estimates of total effects.

Results

Residue Exposure

Data presented in Table 2 are derived from small-scale field testing on alfalfa. RT25 indicates the residual time required to bring bee mortality down to 25% in cage test exposures to field-weathered spray deposits. Materials with an RT25 of 2 h or less can be applied with minimal hazard to bees when they are not actively foraging. Those reaching RT25 within 8 h present a minimal problem to bees, if they are applied during late evening or night. Variations in RT25s are usually associated with low temperatures, since chemicals applied during cool weather retain a longer residual hazard (Johansen 1981). Carbofuran varies from 1 week to greater than 2 weeks of residual hazard which is closely correlated with temperature.

Systemic insecticides also vary in hazard with age of foliage. Table 2 shows an RT25 of less than 1 day for the leafcutting bee when young, succulent alfalfa was treated with methamidophos; while the residual hazard was increased to greater than 5 days when old, senescent foliage was treated.

Nectar Contamination

Only two insecticides are known to have a nectar contamination hazard for bees. Dimethoate at ≥8.9 kg of AI/ha continued to kill alfalfa leafcutting bees for at least 15 days in our greenhouse studies. At the recommended field dosage on alfalfa, 0.45 kg of AI/ha, it has no adverse effects. Waller and Barker (1979) showed that dimethoate was hazardous to honey bees at levels as low as 0.2 ppm when it was fed in sugar syrup to small colonies (ca. 2,000 workers) in caged mini-hives. However, A. Stoner (unpublished data) found that full-sized colonies fed either 0.1- or 1.0-ppm dimethoate performed better than or as well as control colonies in production of sealed brood and maintenance of adult populations. He suggests colonies in the field can tol-

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Table 1. Calibration of residue exposure mortalities and Todd trap catches with expected reduction of honey bee field force and colony

Hazard level	% Mortality 8-h residues	Dead bees/Todd trap per day	Expected % kill: field force	Expected % kill: colony
Very high	90–100	≥3,000	60–100	30–100[a]
High	65–85	1,000–2,000	20–40 (50–100)[b]	10–40
Moderate	35–60	500–900	10–18 (25–45)	5–18
Low	0–30	200–400	4–8 (10–25)	2–8
Normal dieoff	0–5	0–100	0	0

[a] Depends on amount of pollen contamination, length of residual action, and size of colony.
[b] Greater proportion indicated with fast-acting chemical and fewer bees returning to the hives.

erate at least 1-ppm dimethoate because of the diversity of their foraging habits.

Investigators in the Southwest continue to study the repellent and toxic effects to bees of dimethoate applied to lemon or onion.

Aldicarb at 5.36 and 8 kg of AI/ha caused moderate to high mortalities of leafcutting bees for 14 days in greenhouse studies. It caused moderate to high mortalities of alkali bees for 14 days with 2.68, 5.36, and 8 kg of AI/ha. Aldicarb is not hazardous to either bee under recommended field use conditions, since they are not exposed to contaminated nectar until at least 4 weeks after treatment. C. M. Rincker (unpublished data) conducted field cage studies which showed aldicarb has no adverse effects on alfalfa leafcutting bees when the soil applications are properly timed before bloom. No doubt the wild bees' normal tolerance to chemicals safeguards them from aldicarb, because there is no exposure to surface residues on plants with this systemic insecticide injected into the soil. Our acute oral LD_{50}s for aldicarb to alfalfa leafcutting bees, alkali bees and honey bees are 8.9, 4.1, and 0.65 μg/g, respectively.

Contaminated nectar presents minimal, if any, hazard to alkali bee larvae because the stores on which they feed are essentially pure pollen. Our analyses for sugars in freshly prepared pollen balls show they only contain 8.7% nectar.

Pollen Contamination

George and Rincker (1982) consistently found pollen contamination with demeton and nectar contamination with aldicarb, but usually not vice versa. Neither material was detrimental to leafcutting bees in their studies. Arnett (1980) reported unusually high toxaphene residues (up to 56 ppm) in the pollen-nectar stores of leafcutting bees in a certain area of Nevada. Toxaphene was definitely associated with increased levels of immature mortality in this case. Torchio (1983) implicated both trichlorfon and naled as causes of immature mortality in unreplicated field trials. Since a number of other variables could be associated with the small differences in dead eggs and young larvae observed, his results may not be of practical significance. The main factor involved in immature mortality of the leafcutting bee is high temperatures occurring during commercial field operations (Eves and Johansen 1974).

Pollen contamination is the major source of poisoning problems affecting entire honey bee colonies. Pollen-collecting workers moisten their appendages and brush and comb their body hairs to form pellets on the corbiculae. Powder formulations of insecticides are readily incorporated into the pollen loads in this way. Microencapsulated methyl parathion poses a severe hazard because the plastic capsules are about the same size as pollen grains and have a strong affinity for adherence to the bee (Johansen and Kious 1978).

Leaf Piece Contamination

Waller (1969) reported several insecticides with long residual toxicity are hazardous to the leafcutting bee by leaf piece contamination. However, the most damaging materials were DDT and azinphosmethyl, which are no longer used or are not applied to blooming alfalfa. We found leafcutting bee females were killed when they cut leaf pieces from aldicarb-contaminated foliage, but only at excessive dosages and only during the first 10 days after treatment.

Adjuvants

In an effort to obtain greater effectiveness with trichlorfon against lygus bugs, we started testing acidified sprays in 1969 (Johansen and Eves 1972). Using 0.47 to 0.95 liter of a nutrient spray acidifier per 378.5 liters of water typically reduced alkaline waters of about pH 8 down to about pH 6. This level of acidification improves the performance of trichlorfon, but higher levels can greatly increase the hazard to bees.

In Table 3 are mortalities of alfalfa leafcutting bees exposed to 3-h residues of 1.12 kg of trichlorfon (AI) per ha with varying amounts of an acidifier, Sorba-Spray Mg.

We tested foam spray additives and foam spray nozzles and increased the hazard of several insecticides to neither leafcutting bees nor honey bees. Emulsifiable concentrate, soluble powder, and liquid solution formulations of insecticides are much less hazardous to bees than either wettable powder or dust formulations (Johansen and Kleinschmidt 1972). Use of liquid formulations or addition of oily materials to spray mixtures apparently increases sorption of insecticides to plant surface tissues and decreases the hazard to bees (Johansen 1972b).

Table 2. Toxicity of field-weathered residues of insecticides to the alfalfa leafcutting bee (LB), the alkali bee (AB), and the honey bee (HB)

Insecticide	kg of AI/ha	RT25 (h)[a]		
		LB	AB	HB
Acephate	0.45 to 0.9	>72	>72	>72
Azinphosmethyl	0.33	70	—	61
Bromophos	0.9	>24	—	14
Bromophos-ethyl	0.9	>24	—	30
Carbaryl	0.9	70 to >168	70 to >168	70 to >168
Carbaryl (Sevin-4-oil)	0.9	>72	>72	>72
Carbaryl (Sevin-4-oil) ULV	0.45	3	—	2
Carbaryl (Sevin XLR)	0.9	<2	—	<2
Carbofuran	0.9	168 to >336	168 to >336	168 to >336
Carbophenothion	0.9	43	9	<2 to 5
Carbosulfan	0.45	52	—	>72
Chlordimeform	0.9	<2	<2	<2
Chlopyrifos	0.9	>24	>24	>24
Cypermethrin	0.07	>72	—	>72
DDT	1.8	20 to 42	—	<4 to 42
Demeton	0.2	<2	<2	<2
Diazinon	0.9	46	34	49
Dicofol	0.9	<2	—	<2
Dicrotophos	0.45	4 to 66	52 to 96	21 to 37
Diflubenzuron	0.1	<2 to 6	<2 to 6	<2 to 6
Dimethoate	0.45	72 to >72	52 to >72	7 to >72
Dioxathion	0.9	9	—	<2
Disulfoton	0.45	13	<2	<2
Endosulfan	0.45	33 to 80	5	<2 to 3
Endrin	0.9	62	27	<2
Ethion	0.9	<72	—	<3
Etrimfos	0.45	>120	>120	>63
Fenthion ULV	0.9	<2	—	<2
Fenvalerate	0.9	9	7	6
Fluvalinate	0.9	2	26	<2
Formetanate hydrochloride	0.9	4 to 14	3 to 9	<2 to 2
Isofenphos	0.9	>24	>24	>24
Isopropyl parathion	0.45	<2	<2	<2
Leptophos	0.9	27 to 30	2 to 16	<2 to 3
Malathion	0.9	57	—	40
Malathion ULV	0.56	158	—	131
Menazon	0.9	<2	—	<2
Methamidophos	0.9	22 to >120	19 to 102	7 to 25
Methidathion	0.9	12 to 64	13 to 74	24 to 80
Methomyl	0.4	6 to 15	5 to 8	2
Methoxychlor	2.7	2 to 4	<2	<2
Methyl parathion	0.45	22	21	20
Mevinphos	0.45	<5	<5	<5
Monocrotophos	0.45	>24	>24	>24
Naled	0.9	>72	20 to 46	12 to 20
Oxamyl	0.9	54	47	12
Oxydemetonmethyl	0.45	<2	<2	<2
Parathion	0.45	15 to 64	21 to 27	13 to 18
Permethrin	0.9	>48	32 to 42	14 to 42
Phenthoate	0.9	>24	>24	>24
Phosalone	1.34	<2	<2	<2
Phosmet	0.9	>24	>24	>24
Phosphamidon	0.45	52	25 to 46	16 to 40
Phostex	0.9	<2	—	<2
Phoxim	1.34	>48	37	39
Pirimicarb	0.2	<2	<2	<2
Pirimiphos-ethyl	0.9	>48	—	29
Pirimiphos-methyl	0.45	9	—	7
Profenofos	0.9	>24	>24	9
Propargite	1.3 to 1.8	<2	<2	<2
Propoxur ULV	0.14	<2	—	<2
Ronnel	0.9	70	—	30
Schradan	0.9	<2	<2	<2
Temephos	0.5	2 to 40	<2 to 15	<2
TEPP	0.45	<5	<5	<5
Tetrachlorvinphos	0.9	24	10	<2
Tetradifon	0.7	<3	<2	<2
Thiodicarb	0.9	8	—	<2
Toxaphene	2.7	78 to 168	10	<2 to 4
Toxaphene + DDT	1.3 + 2.7	27 to 95	13 to 52	8 to 32
Trichlorfon	0.9	<2 to 5	6 to 14	<3 to 6

[a]RT25 indicates the residual time required to bring bee mortality down to 25% in cage test exposures to field-weathered spray deposits.

Table 3. Mortalities of alfalfa leafcutting bees (LB) exposed to 3-h residues of 1.12 kg (AI) of trichlorfon per ha

Liters of acidifier/378.5 liters	LB % mortality
0	34
0.95	62
1.9	71
3.8	87

Table 4. Mortalities of alkali bees (AB) exposed to 2-h residues of propargite and propargite-insecticide mixtures

Treatment	kg (AI)/ha	AB % mortalities
Propargite	1.8	0
Trichlorfon + demeton	0.9 + 0.2	17
Propargite + trichlorfon	1.8 + 0.9	22
Propargite + trichlorfon + demeton	1.8 + 0.9 + 0.2	50

A new formulation of carbaryl, Sevin XLR, is much lower in hazard to bees than all previous flowable formulations of this chemical. Use of a much smaller particle size and addition of a latex sticker contribute about equally to the safening process. Recent tests of a proprietary sticker in combination with an acephate soluble powder gave similar promising results in lowered toxicity to bees.

Synergism

When dicofol, tetradifon, or propargite are added to one or more insecticides in spray mixtures, they greatly increase the toxic hazard to bees. Such action appears to be true synergism, since the specific acaricides are essentially nontoxic when used alone.

Percent mortalities of alkali bees exposed to 2-h residues of propargite and propargite-insecticide mixtures were as shown in Table 4.

Age Effect

After leafcutting bee females have been actively nesting in the field for 3 or more weeks, they become much more susceptible to insecticides. We have shown this with trichlorfon, formetanate hydrochloride and naled and, not surprisingly, the untreated checks also increase in mortality with age of bee.

In Table 5 are the mortalities of three age groups of alfalfa leafcutting bees exposed to 4-h residues of several insecticides.

Herbicides and Fungicides

Most herbicides tested have proven to be essentially innocuous to honey bees, alkali bees, and alfalfa leafcutting bees. The most notable exceptions which are hazardous to bees are arsenicals, endothal, and DNBP (dinoseb). Even the high-dosage levels of soil residual herbicides used for vegetation management on alkali bee soil nesting sites are nonhazardous to both adults and larvae (Johansen et al. 1978).

Table 5. Mortalities of three groups of alfalfa leafcutting bees (LB) exposed to 4-h residues of insecticides

Treatment	kg (AI)/ha	LB mortality at the following ages (wk)s		
		0	3	6
Trichlorfon	0.9	5	38	61
Formetanate hydrochloride	0.4	27	74	100
Naled	0.9	91	100	100
Untreated check	—	7	10	22

All fungicides tested have proven to be essentially nontoxic to bees, even when applied directly to them.

Poisoning Symptoms

Whereas honey bees slow down, lose their ability to fly, and act almost as if they are chilled when poisoned by carbaryl, alfalfa leafcutting bees become very agitated, fly hectically, and often spin on their backs. They exhibit similar symptoms when poisoned by aldicarb, but honey bees also become highly agitated in this case. Alkali bees tend to perform numerous, rapid turning motions while lying on their sides after exposure to aldicarb.

Another fairly typical symptom of poisoning in leafcutting bees and alkali bees is simply a lack of nesting females in field shelters and at soil nesting sites. Large masses of dead and dying bees in front of the hives, which are the commonest sign of poisoning in honey bees, are seldom seen with either leafcutting bees or alkali bees.

Some persons have associated a lack of carrying either pollen or leaf pieces by leafcutting bee females or pollen by alkali bee females with insecticide poisoning. However, we believe this behavior is caused by old age or senility, since it is observed beginning about 4 weeks after the start of female nesting activity in the field regardless of any insecticide exposure.

Discussion

Many factors affect the hazard of pesticides to bees. In addition, alkali bees, honey bees and alfalfa leafcutting bees often react differently following exposure to pesticides. Following are major points of comparison we have made about these interactions.

(1) Alfalfa leafcutting bees are inherently more tolerant to insecticides than the other two species as shown by topical drop and acute oral studies. Alkali bees are intermediate in inherent tolerance to insecticides.

(2) Alfalfa leafcutting bees are typically more susceptible to insecticides than the other two species in the field because they have a higher surface-to-volume ratio, allowing more efficient accumulation of lethal dosages. Alkali bees are intermediate in susceptibility to field-weathered residues of insecticides.

(3) Residual hazard of insecticides to bees is increased with cool temperatures.

(4) Systemic insecticides retain a longer hazard when old, senescent foliage is treated.

(5) Aldicarb and dimethoate are the only two insecticides presently known to have a nectar-contamination hazard to foraging bees. However, neither material presents a problem when properly applied in the Pacific Northwest.

(6) Toxaphene is the only insecticide which has been positively associated with mortality of immature alfalfa leafcutting bees because of contamination of the pollen-nectar stores.

(7) Insecticide contamination of the pollen loads of foraging honey bee workers is the major source of hive contamination and destruction of colonies.

(8) Long surface-residual insecticides such as DDT and azinphosmethyl have a leaf-piece contamination hazard to alfalfa leafcutting bee females.

(9) Acidifiers used at less than 1:400 dilution increase the hazard of materials such as trichlorfon to all three species of bees.

(10) Use of liquid formulations, addition of oily materials or stickers, and use of smaller particle sizes in flowable formulations tend to make insecticides safer to bees.

(11) Acaricides have a synergistic action when added to insecticide sprays which increases the hazard of such mixtures to bees.

(12) Alfalfa leafcutting bees become more susceptible to insecticides with increasing length of activity in the field.

(13) All fungicides and most herbicides have proven to be of little or no hazard to bees to date.

(14) Whereas honey bees become relatively inactive and unable to fly after exposure to carbaryl; alfalfa leafcutting bees and alkali bees become highly agitated and move erratically.

(15) The common symptom of large masses of dead and dying honey bees in front of the hives seldom occurs with the wild bees. Most commonly, there is simply a lack of alfalfa leafcutting bee or alkali bee females at the nesting sites.

(16) Residue exposure mortalities and Todd trap catches can be related to expected kill of the honey bee field force and of the colony.

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